

Deoxynivalenol (DON)

Deoxynivalenol (DON) is a mycotoxin produced by *Fusarium* species, which infect numerous grains, both in the field and during storage. DON was nominated because of its inherent toxicity and potential for widespread human exposure. The primary route of DON exposure is consumption of contaminated grains. In humans, DON is known to cause acute toxicity including headache, dizziness, nausea, vomiting, and diarrhea. In animals, short term exposure results in anorexia and vomiting while long term exposure results in effects on several organs. DON is also known to be genotoxic, immunosuppressive, teratogenic, and it affects multiple reproductive endpoints.

To add to the already well-characterized toxicity profile of DON, NTP has proposed “definitive” long-term studies to address reproductive toxicity, chronic toxicity, and genotoxicity, despite abundant existing data for several of these endpoints from DON as well as data from related trichothecene mycotoxins. Therefore, we question the criterion for “definitive” – what will make the studies proposed by NTP more definitive than the weight of evidence of the existing data?

Furthermore, we question the utility of this data for protecting public health. None of the testing proposed will generate data that will provide for anything beyond refinement of existing regulatory limits. Numerous regulatory authorities have already established maximum daily intake values and set limits for DON in grains used for human and animal consumption. Additionally, in the US, the USDA as well as producers, millers, and processors monitor for the presence of DON in order to prevent contaminated grains from entering the food supply. The NTP Research Concept document states that the possibility of high levels of DON entering the food supply is likely to have been substantially decreased over the last 10 years. The Chemical Information Review Document goes so far as to state that “DON does not pose a threat to public health among the general population”. This raises an important question – what percentage of grain is currently contaminated, at what levels, and do these levels fall within the range of concern? From a risk assessment standpoint, if the levels detected are low and the risk is low, then that should call into question the value of new animal tests. The small, incremental benefit to either scientific knowledge or public health provided by further toxicity testing of DON is outweighed by the costs in terms of money, time, and the suffering inherent in these studies.

The most appropriate risk management action to protect the public from DON is to reduce exposure by reducing the amount of contaminated grains entering the food supply. Existing analytical chemistry, PCR, ELISA, and biosensor based methods for DON screening can be applied on a wider scale to accomplish this goal.

If more data on the effects of DON is perceived to be required, then the focus should be on developing a more accurate understating of the relationship between DON intake and toxic effects in humans through epidemiological studies in areas where *Fusarium* contamination is prevalent. Such information would be more directly relevant than “clarification of species differences” – an exercise proposed in the test plan which includes toxicokinetic studies in both rats and mice.

We also disagree with the proposed prechronic rat study for development of toxic equivalency factors (TEFs) and dose selection for reproductive and carcinogenicity studies. With regard to dose selection, we would ask NTP to consider using dose information from the myriad existing studies rather than conducting new testing for this purpose. NTP is also interested in developing TEFs for evaluation of cumulative risk, but we don’t understand why a rat study is necessary for reaching that goal. A potentially more relevant approach for assessing cumulative risk would be to evaluate the relative toxicity of the various trichothecene mycotoxins *in vitro* where any number of toxicity endpoints and pathways could be assessed in cell types from multiple species including humans. Perhaps some of the assays of the High Throughput Screening Initiative of the Biomolecular Screening Branch at NTP

could be used to determine the relative toxicities of the trichothecene mycotoxins for endpoints of interest including carcinogenicity, reproductive toxicity, and genotoxicity. This approach should be more than adequate for generating a potency scale and could be a powerful tool for determining which toxins add to the total toxicity and by how much, thereby facilitating assessment of cumulative exposure.

The proposal to perform a reproductive toxicity assay in rats is surprising given the availability of substantial existing data relevant to this endpoint. There are numerous studies in rats, mice and pigs with DON and related *Fusarium* mycotoxins indicating various reproductive effects in both males and females ranging from abnormal sperm to decreased fertility and number of live pups per litter. These studies were sufficient to establish NOELs and LOELs, therefore, given all of this evidence, we ask that the proposed reproductive studies be reconsidered and we question how another study will provide more definitive data than what already exists. Additionally, NTP repeatedly cites the Joint FAO/WHO Expert Committee on Food Additives and the European Commission Scientific Committee on Food (JECFA) findings on DON to support NTP's proposed testing, but in its exhaustive review of DON, JECFA's final recommendations for further work do not include additional reproductive testing. In fact, the JECFA committee established a provisional maximum tolerable daily intake of 1 µg/kg bw, stating, "that intake at this level would not result in effects of deoxynivalenol on the immune system, growth, or reproduction."

In closing, we ask that NTP reconsider the animal toxicity testing proposed. We believe that resources would be better spent on exposure assessments and measures that could more directly protect public health. In its final recommendations, JECFA has identified several areas in this vein that require more work, which include: conducting more detailed, human studies; gathering better data on the distribution of DON contamination; developing better methods of measurement for DON in processed foods; better data on food consumption patterns; and better tools for the prevention of *Fusarium* infection of grain.